with a heterogenous assemblage of species, and *P. gracilis* and *P. hagenianus* in the *Gracilis* section. Hobbs (1942) revised this classification and placed the three species mentioned above in a separate group (the *Simulans* group) of his newly established *Barbatus* Section. *Procambarus tulanei* clearly belongs to the *Simulans* group and is most closely related to *P. simulans*. *P. tulanei* and *P. simulans* closely resemble each other in most superficial characteristics. The two species can be separated easily by the structure of the first pleopod of the male (*cf.* Figs. 9 and 13), but not by the annuli ventrali of the females which are quite variable in both species. The geographic ranges of these two species meet in western Louisiana but apparently do not overlap.

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HELMINTHOLOGY.—*Studies on the helminth fauna of Alaska: XII, The experimental infection of Alaskan gulls* (Larus glaucescens *Naumann*) *with* Diphyllobothrium *sp.*

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For over a period of three years cestodes belonging to the genus *Diphyllobothrium* Cobbold (1858) have been routinely recovered from various Alaskan mammals, including man. The epidemiological nature of this helminthic infection is not yet completely understood. Experimental as well as field research has been undertaken by the Animal-borne Disease Branch of the Arctic Health Research Center relative to the possible control of diphyllobothriasis in Alaska. The results of the work to date, including the laboratory infection of dogs (*Canis familiaris*), foxes (*Vulpes fulva* and *Alopex lagopus*), bears (*Ursus americanus*), gulls (*Larus glaucescens*), and man, present strong evidence that only one species is involved. However, since its life cycle has not been experimentally determined, and since morphological study does not allow speciation of the adult worm, this must remain only an assumption. The present paper on the infection of gulls with *Diphyllobothrium* sp. constitutes a preliminary report.

The writer wishes to express his gratitude to Dr. Robert Rausch, chief of the Animal-borne Disease Branch, under whose supervision this work was carried out and who identified the cestode material, and to Miss Reggie Sacressen, whose aid in connection with this project contributed greatly to its success.

MATERIAL AND METHODS

For these experimental infections young gulls (*Larus glaucescens*) were secured from isolated nesting grounds on islands in Cook Inlet, about 20 miles southwest of the city of Anchorage. The numerous small island nesting sites were heavily grown to sedges and thus afforded ample protection for the young of hundreds of birds, among which several species were represented.

Thirty-six birds taken on June 21, 1951, were downy young; these were collected directly from their nests, or in the immediate vicinity. The nests were numerous and widely scattered and, in several instances, still contained unhatched eggs. The estimated ages of the birds taken ranged from 1 day to 2 weeks, and their weights were from 55 to 340 grams.

On July 15, 15 juvenile birds were collected from these same nesting grounds. The young gulls were located by flushing them from their grassy niches and by cutting off their retreat as they swam to shore. At this time it was noted that a few nests still contained eggs; examination of several of these revealed that they contained dead embryos. The young gulls taken were apparently from 1 to 6 weeks old and weighed 207 to 1,034 grams.

As a control 22 of the first group of 36 birds taken on June 21, and all those collected July 15, were examined post mortem, at the time of collection, to determine the extent of natural helminth infections. No parasites were recovered from the downy-young birds, and only one species

of a strigeid trematode was taken from the juveniles.

The 14 gulls intended for experimental use, after having been observed for about two weeks and having become accustomed to captivity, were color-banded, divided into five groups (see Table 1), and placed in five separate cages—designated A, B, C, D, and E.

During the course of the experiment, king salmon (*Oncorhynchus tshawytscha*), which had been quick-frozen and kept for over a year at a temperature range of −12° to −16° C., was used for feeding. At the time of collection it was noticed that the young birds were being fed exclusively on salmon brought to them from the beaches or from the refuse dumps of nearby canneries. Since there were no lakes nearby where infected fishes could be obtained, this seemed to preclude preinfection by *Diphyllobothrium* sp. The literature does not report, so far as the writer is aware, that this fish species harbors the plerocercoids of any species of *Diphyllobothrium*; consequently, the feeding of salmon by the gulls to their young would not seem to have any bearing on this experiment. The fact that our salmon had been stored for such a length of time at low temperatures made it highly improbable that larval cysts, if present, could still be viable.[1] During the first week of captivity, the young gulls were fed small pieces of salmon every three hours. The frequency of feeding was reduced with the age and the development of the birds.

Plerocercoids for the experiment (muscular and visceral in location) were recovered from rainbow trout, *Salmo gairdnerii* Richardson, taken from Daniels Lake on the Kenai Peninsula. The fishes of this lake had been previously studied by us and were known to be infected with *Diphyllobothrium* sp.[2] On July 7 viable plerocercoids were fed to the young gulls as follows: Each of the birds in cages A and B (see Table 1) received five visceral cysts and those in cages D and E received muscular cysts—three and four larvae each, respectively. The third bird of cage E (no. 14), after having seized some infected fish which was not intended for him, was rewarded with the entire fish; thus an unknown number of larvae was given in this case. On July 15 the experimental feeding of the birds in cages A and D was repeated, with each receiving the same number of larvae as before. At this time, both of the gulls in cage C were infected with five visceral cysts, making a total of 14 artificially infected birds. Autopsy of the young gulls was begun on the second day after the last infection and continued through the twenty-fourth day (see Table 1).

[1] Kjava (1913), as quoted in Magath and Essex (1931) stated that submitting infected fish to a temperature of −9° C. for 24 to 48 hours kills the larvae of *D. latum*; this was confirmed by the latter authors.

[2] This had been determined by morphological study of adult worms obtained through feeding plerocercoids to certain mammalian hosts.

RESULTS AND DISCUSSION

As may be noted from the table, seven gulls, or 50 percent, were found to be infected with *Diphyllobothrium* sp.; this included two of the eight birds that had received visceral cysts and five of the six that had received muscular cysts. It is probably incidental that those receiving muscular cysts showed a higher degree of infection. None of the birds in cage A was infected, despite their having received the most larvae. The small number of birds involved does not, however, permit any conclusions. Segment counts of three 13-day-old strobilae gave an average of 333. This figure may serve to indicate the rapid rate

Table 1—Experimental Infection of Gulls with Diphyllobothrium sp.[1]

Host No.	Date of infection		Number of larvae administered	Larval type		Days elapsing before autopsy	Number of worms recovered
	July 7	July 15		Muscle	Visceral		
Cage A							
1	X	X	10		X	2	0
2	X	X	10		X	6	0
3	X	X	10		X	24	0
Cage B							
4	X		5		X	2	0
5	X		5		X	6	0
6	X		5		X	13	0
Cage C							
7		X	5		X	7	2
8		X	5		X	12	4
Cage D							
9	X	X	6	X		2	3
10	X	X	6	X		6	3
11	X	X	6	X		24	1
Cage E							
12	X		8	X		2	6
13	X		8	X		6	0
14	X		?	X	X	13	37

[1] Thirty-seven controls, negative for helminth parasites, are not included in table.

of growth. All worms recovered were fixed, stained, and mounted for morphological study.

Macroscopic examination of the specimens of *Diphyllobothrium* sp. recovered from artificially infected gulls, as well as those from dogs, foxes, bears, and man, reveals a great range of variation in gross appearance. Possibly the host species involved, its physiological condition, and the age of the worm may be the primary determining factors in the morphological variation of the adult form. Stunkard (1949) pointed out that "species now included in the genus constitute a heterogeneous collection, from a variety of hosts and bionomic areas, but the morphological diversity is so distributed among the species and so many different combinations of characters exist that the arrangement of the species into related groups must await further information." Rausch (1951) stated, "Speciation on the basis of morphological characters of tapeworms of the genus *Diphyllobothrium* is impossible for the Alaskan forms." These statements indicate that a new approach, rather than a taxonomic one, must be made to the problem of speciation of the worms of this genus.

The successful infection of gulls with *Diphyllobothrium* sp. considerably complicates the problem of control of this helminth in Alaska. Bent (1921), in connection with the glaucous-winged gull, wrote, "This is the most abundant, the most widely distributed, and characteristic gull of the north Pacific coast . . ." The vast numbers of these birds, coupled with their potential for disseminating viable eggs over a wide area will undoubtedly enhance the chances of survival of this parasite in Alaska. Terrestrial mammals which utilize fishes for food also constitute an important reservoir of infection, but nevertheless this is one which probably could be controlled locally if diphyllobothriasis were ever thought important enough to human health to justify extreme measures.

The low living standard of the Alaskan native often compels him to depend on fish almost exclusively or at least as a substantial part of his diet. Local customs and the conditions under which many of these people live necessitate the eating of raw or poorly cooked fish. Preliminary surveys made to date reveal that the percentage-incidence of infection among the natives in certain areas where *Diphyllobothrium* sp. is endemic is quite high. Hitchcock[3] (1950; 1951), during her parasitological investigations of the Bethel and Kotzebue areas, found an incidence of infection of 15 and 6 percent, respectively. Although the pathogenicity of *Diphyllobothrium* is not well understood, it seems probable that the disease may aggravate, in areas where it is endemic, the present serious health condition of Alaskan natives. Consequently, diphyllobothriasis must be regarded as a definite public health problem in Alaska.

A more intensified investigation of the immunological characteristics of this parasite in native birds and mammals in Alaska will be undertaken during the field season of 1952.

SUMMARY

The experimental infection of glaucous-winged gulls (*Larus glaucescens*) with *Diphyllobothrium* sp. has been accomplished. All available evidence indicates that this Alaskan species readily infects gulls, various wild carnivorous mammals, and man. The specific identity of this cestode has not been determined.

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[3] Miss Hitchcock was employed as a consultant for the Arctic Health Research Center at the time these surveys were made.